Nanotrap Extracellular Vesicle B Particles Protocol

This protocol describes a method using magnetic Nanotrap® Extracellular Vesicle B Particles to concentrate intact extracellular vesicles that were isolated using size exclusion columns, such as the qEV columns from Izon Science Limited.

Contents and Equipment Lists

Download the Kit Contents and Equipment Lists PDF at https://www.ceresnano.com/ev/ev-user-guide.

Troubleshooting

- Ensure that all reagents are stored at the indicated temperatures.
- Make sure refrigerated reagents and samples (if applicable) are warmed to room temperature before starting the procedure.
- If you are interested in exploring different applications for the Nanotrap® Extracellular Vesicle Particles, please contact support@ceresnano.com for technical assistance.
- Nanotrap Extracellular Vesicle B Particles must be adequately vortexed immediately before each use.
- Nanotrap Extracellular Vesicle B Particles are not suitable for use in conjunction with RNA extraction kits that use alcohols in the lysis buffer, as it decreases the lysis efficiency of extracellular vesicles that are bound to the Nanotrap Extracellular Vesicle B Particles. Please ensure any RNA extraction kit used downstream is compatible with this kit before use. RNA extraction kits can be used if the lysis buffer is substituted for one that does not contain alcohols, such as MagMAX™ Microbiome Lysis Solution, (part number A42361). This is a guanidine thiocyanate based lysis solution that does not contain alcohol.
- Once the Nanotrap Extracellular Vesicle B Particles have been pelleted, the lysate can then be removed and used in any RNA extraction protocol.
- Note that extracellular vesicles bind irreversibly to Nanotrap Extracellular Vesicle B Particles. This is an important consideration for extracellular vesicle quantification analysis and functional studies downstream.
- After following the protocol, if there are still extracellular vesicles present in the supernatant that have not bound to the Nanotrap Extracellular Vesicle B Particles, add a fresh aliquot of Nanotrap Extracellular Vesicle B Particles (the same volume as specified in the table) to the supernatant and repeat the incubation and magnetic separation steps.

Guidelines

After the isolation of extracellular vesicles using a size exclusion column, follow the steps below to concentrate the extracellular vesicles. The volume of Nanotrap Extracellular Vesicle B Particles required to concentrate extracellular vesicle-containing samples is dependent on the volume of the purified extracellular vesicle sample. See **Table 1** for suggested volumes of Nanotrap Particles to be used for different volumes of purified extracellular vesicles. If you are unsure how to prepare your sample or what volume of Nanotrap Particles to add, please contact support@ceresnano.com for assistance.

Table 1: Recommended volume of Nanotrap Extracellular Vesicle B Particles to be added to purified extracellular vesicle samples

Volume of Purified Extracellular Vesicles	Volume of Nanotrap Extracellular Vesicle B Particles
0.51 – 0.85 mL	20 µL
1.2 – 2.0 mL	40 µL
2.1 – 3.5 mL	70 µL

Concentrating Extracellular Vesicles

In a clean lo-bind tube of the appropriate size, complete the following steps, in order.

1. Add the sample containing the purified extracellular vesicles to the sample tube.
2. Vortex Nanotrap Extracellular Vesicle B Particles for 30 seconds to resuspend.
3. Add the appropriate volume of Nanotrap Extracellular Vesicle B Particles **(see Table 1)** to the sample tube.
4. Vortex sample tube for 30 seconds to ensure that the Nanotrap Extracellular Vesicle B Particles are fully resuspended and incubate for 10 minutes at room on a tube roller or inverter.
5. Place the sample tube on the magnetic rack for 2 minutes to pellet the Nanotrap Extracellular Vesicle B Particles.
6. While on the magnetic rack, remove the supernatant, being careful not to disturb the pellet containing extracellular vesicles bound to Nanotrap Extracellular Vesicle B Particles.
7. The extracellular vesicle pellet is now ready for downstream applications. The pellet can be resuspended in a desired buffer volume or used directly with method-appropriate lysis buffer, to avoid further dilution of concentrated extracellular vesicles.

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